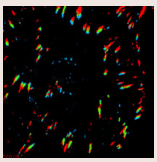
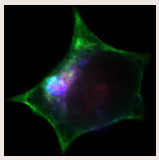
In this issue



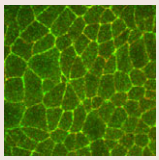
Focal adhesions feel the force

Focal adhesions are dynamic mechanosensitive structures that can respond to changes in internal and external forces, but inhibiting actomyosin contractility results in the breaking down of focal adhesions and prevents the formation of new ones. Benjamin Geiger and co-workers (p. 1425) now shed light on the mechanisms that underlie this mechanosensitivity. The researchers use high temporal resolution FRAP and direct measurement of focal adhesion protein dissociation to analyse the effect that inhibiting actomyosin-generated forces with blebbistatin has on the molecular make-up of focal adhesions. They find that a key determinant of the turnover of focal adhesions is the dissociation rate constant (k_{off}) of individual proteins in the complex, which is sensitive to changes in applied force. Geiger and co-workers further describe that reduced tension leads to distinct effects on the turnover of three focal adhesion proteins: vinculin, paxillin and zyxin. Whereas inhibition of myosin II with blebbistatin results in increased dissociation of vinculin from the adhesions, the dissociation of paxillin and, to a lesser extent, zyxin from focal adhesions is attenuated. The authors propose that it is these changes in molecular kinetics that lead to changes in focal adhesion turnover, composition and, ultimately, their disassembly under conditions of reduced tension.



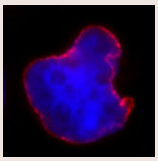
Aggregates: CK2 to the rescue

Misfolded proteins are bad news for cells because they can easily form aggregates that interfere with normal cellular functions. Usually, such accumulation is carefully avoided by degradation of misfolded proteins by the proteasome. If this pathway becomes impaired or overloaded, however, the aggresome provides a second line of defence: misfolded protein aggregates are transported to this inclusion-body-like organelle by the deacetylase HDAC6 and dynein motors along the microtubule network, and are later removed by autophagic degradation. Although it is well known that the aggresome has evolved to cope with an excess of protein aggregates, the mechanisms underlying its formation have remained unclear. On page 1519, Masahiko Watabe and Toshio Nakaki now identify the protein kinase CK2 as a crucial factor in aggresome assembly and clearance. They show that CK2 provides a linker that facilitates the interaction between HDAC6 and dynein. Furthermore, in response to cellular stress caused by misfolded proteins, the kinase phosphorylates HDAC6 on serine 458. The resultant increase in HDAC6 deacetylase activity is crucial for both the recruitment of misfolded proteins to, and their clearance from, the aggresome. CK2 thus has an important role in maintaining cell viability in times of increased stress from misfolded protein aggregrates.



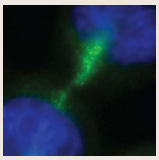
APC2 links ARMs to organise actin

Adenomatous polyposis coli (APC) proteins localise in the cytoplasm, at the cell cortex and in the nucleus, where they coordinate the negative regulation of Wnt signalling and direct actin and microtubule organisation and function. The mechanisms that determine APC localisation, and how this localisation contributes to the cellular roles of these proteins, however, remain poorly understood. On page 1589, Brooke McCartney and colleagues address these longstanding questions, first by demonstrating that two domains of *Drosophila melanogaster* APC2 are required for cell cortex localisation, and second by characterising the subsequent impact of this localisation on both Wnt signalling and actin organisation. The authors show that the localisation of APC2 to the cell cortex is dependent on both the ARM repeats, which undergo self-oligomerisation to form higher-order complexes of APC2, and a new domain that contains the C-terminal 30 amino acid residues. The authors propose that APC2 ARM oligomerisation might be required to bring multiple C-terminal domains into close proximity, which could provide a docking site for a protein that would link APC2 to cortical actin. Interestingly, the authors demonstrate that this C-terminal domain – and indeed, APC2 cortical localisation – is essential for actin organisation in the *Drosophila* embryo, but not for regulation of Wnt signalling.



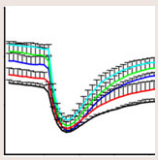
Nuclear protein 4.1R gets starring role

The nucleus – arguably the cell's most important organelle – is anything but simple. In addition to housing DNA and all of its associated protein machinery, the nucleus also contains an extensive network of structural proteins. One of these is called protein 4.1R, which was first described as an essential mediator of erythrocyte shape and mechanical stability. Protein 4.1R has since also been described in nucleated cells, where it is crucial for the assembly and maintenance of functional nuclei. 4.1R acts as a linker protein that interacts with diverse binding partners. But which interactions are key to its roles in regulating nuclear structure and function? On page 1433, Sharon Wald Krauss and colleagues now provide an answer to this question by showing that 4.1R associates with the nuclear envelope protein emerin and the intermediate filament protein lamin A/C. Furthermore, depletion of 4.1R perturbs lamin A networks, emerin localisation and the distribution of other nuclear proteins. These changes impact upon nuclear morphology, and the distance between the centrosome and the nuclear envelope, and cause an increase in nuclear β-catenin transcriptional activity. In mice, the partial knockdown of 4.1R results in disease phenotypes, such as cardiac and kidney defects and neurobehavioural deficits. Thus, 4.1R is clearly a key player that links nuclear and cytoplasmic structures to maintain nuclear shape and function.



Buckle and fuse to divide

Abscission is the final event in cytokinesis, separating one daughter cell from the other. During cytokinesis, the contractile actomyosin ring constricts the plasma membrane at the division site and compacts the central spindle microtubules within the intracellular bridge (ICB). To allow fusion of the plasma membrane, these microtubules have to be disassembled – but how is this achieved? Using a combination of time-lapse microscopy and high-resolution 3D tomography, Rytis Prekeris and colleagues (p. 1411) now uncover the mechanisms that mediate abscission. The authors demonstrate that localised depolymerisation of the central spindle microtubules, which comprise most of the ICB, is not achieved by spastin-mediated severing, as originally thought, but by microtubule buckling. This buckling, and subsequent breaking, of the microtubules frees space in the ICB, so that FIP3- and VAMP8-containing recycling endosomes can be recruited and fuse with the plasma membrane in the next step of abscission. The increased rate of organelle fusion, in turn, enhances plasma membrane dynamics in the ICB and thereby leads to the formation of a 'secondary ingression' – a crucial step that drives the thinning of the ICB and, finally, scission. With these results, the researchers show that buckling of microtubules in the ICB is a key event in the process of abscission.



PECAM-1 maintains the barrier

Platelet endothelial cell adhesion molecule (PECAM)-1 is highly expressed at endothelial cell–cell junctions in confluent vascular monolayers and is necessary for vascular barrier integrity. PECAM-1 is both a cellular adhesion receptor and a signalling receptor – yet which, if any, of these properties is behind PECAM-1-mediated barrier protection? Here, Jamie Privratsky, Peter Newman and co-workers (p. 1477) compare primary human endothelial cells expressing various PECAM-1 variants in order to answer this question. As expected, PECAM-1-expressing endothelial cells have a higher level of barrier integrity when compared with those deficient in PECAM-1. Interestingly, the authors found that this higher level of protection is dependent on PECAM-1–PECAM-1 homophilic interactions between the PECAM-1 extracellular domains; this homophilic interaction is required to direct PECAM-1 to cell–cell junctions. However, mutations of residues in PECAM-1 that are known to be essential for signal transduction, including conserved immunoreceptor-based tyrosine motifs (ITIMs) and residues shown to direct PECAM-1 recruitment to membrane microdomains, have limited impact upon barrier integrity. The authors suggest these results mean that the cell adhesion role of PECAM-1 is more important for maintaining barrier integrity than its signalling function.